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COMBINED EFFECTS OF TEMPERATURE AND SALINITY ON EGG HATCHING RATE AND INCUBATION TIME OF *PENAEUS SEMISULCATUS* (DECAPODA: PENAEIDAE)

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Abstract

This study was undertaken to determine the combined effects of temperature and salinity on the incubation time and hatching rate of *Penaeus semisulcatus* eggs in controlled laboratory conditions. Fertilized eggs, from a female caught in nature and spawned in our laboratory, were stocked in 2-l round bottom glass flasks and received one of nine temperature (24, 28 and 32°C) and salinity (30, 35 and 40 ppt) combinations. The eggs hatched in all treatments. The hatching rate increased as salinity increased and was highest at 24°C. In terms of hatching rate, the best combinations in descending order were 24°C at 40 ppt, 32°C at 40 ppt and 28°C at 40 ppt. However, the incubation time was longer at 24°C (17.5 h) than at 28°C (14.5 h) or 32°C (11.5 h).

Introduction

Optimum egg incubation conditions are important for hatchery success. Salinity and temperature are two of the most important environmental factors affecting the hatching rate and time of eggs. In nature, spawning, hatch-

ing and larval stages of penaeid shrimps generally take place in oceanic sea waters where salinity and temperature do not fluctuate considerably. It is commonly accepted that penaeid eggs hatch between 12 and 17 h at

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oceanic salinities and 25–28°C. However, little is known about the combined effects of temperature and salinity on the hatching rate and time of penaeid eggs. The optimal temperature and salinity combination for egg incubation is species-specific (Preston, 1985) and these levels must be determined for each species.

A limited number of studies have focused on the effects of salinity and temperature separately. Adequate egg incubation temperature for *P. monodon* was suggested to be 26–29°C (Hillier, 1984; Primavera, 1985). Primavera (1985) recorded that an increase in temperature from 23°C to 28°C or 33°C significantly decreased the incubation period but had no effect on the hatching rate. This author reported high hatching rates at 33 ppt between 23 and 33°C but weak larval activity after hatching at 23 ppt regardless of the experimental temperature. The eggs of *Fenneropenaeus indicus* and *P. semisulcatus* at 20–25 ppt exhibited retarded development and even burst at 10–15 ppt (Tseng and Cheng, 1981; cited in Primavera, 1985). Nisa and Ahmed (2000) found that the best egg hatching rate of *F. merguensis*, *F. penicillatus*, *Metapenaeus affinis* and *Parapenaeus stylifera* took place at 35 ppt, and that the hatching rate decreased gradually with decreasing salinity until no hatching occurred at 20–25 ppt for *P. stylifera*. Most recently, Zacharia and Kakati (in press) studied the combined effects of temperature and salinity on the hatching success of *F. merguensis* and found that the highest percent hatch was obtained at 33°C and 35 ppt (87%), followed by 29°C and 35 ppt (82%).

The hatching rate of eggs of eyestalk-ablated females of *P. semisulcatus* was 47.82% at 38 ppt and 28°C (Aktas and Kumlu, 1999) and 90.3% at 22.4–27.5°C (Browdy and Samocha, 1985). To date, no studies have been carried out to determine the optimum combination of salinity and temperature for *P. semisulcatus*, an Indo-Pacific species distributed along the coast of the eastern Mediterranean and one of the most important commercial species in this region. Kumlu et al. (1999a) observed that the spawning season of *P. semisulcatus* extends into cool

months, hence this species might have adapted to subtropical conditions. A better understanding of the combined effects of temperature and salinity on the hatching rate and incubation time for eggs of this penaeid shrimp is important to determine conditions for optimal hatchery production. Therefore, this study was carried out to determine the salinity and temperature combination that produces the best hatching rate and shortest egg incubation time for *P. semisulcatus*.

Materials and Methods

The study was undertaken at the Marine Research Station of the Faculty of Fisheries, University of Çukurova, Yumurtalık, Turkey. The eggs were obtained from a female caught off Yumurtalık Bight in the northeastern Mediterranean, that matured in the station and spawned in a 100-l tank at 39 ppt and 28°C. The fertilization rate was determined under a microscope. Ten minutes after spawning, the eggs were pooled in a 100-µm sieve and acclimated to the experimental salinities (30, 35 and 40 ppt) and temperatures (24, 28 and 32°C) by lowering or increasing the salinity/temperature at a rate of 2 ppt or 1°C per 15 min, respectively. Salinity was adjusted by adding fresh water (well water) that was prepared and aerated before use or sea salt (Instant Ocean, USA) as described in Kumlu et al. (2000, 2001). The eggs were stocked into 2-l round bottom glass flasks in three replicates of each combination at a density of 50 eggs per liter.

Salinity and temperature were measured with a digital salinometer (YSI 30, USA). Temperatures for each treatment were maintained in three thermostatically controlled water baths ($\pm 0.5^\circ\text{C}$). Gentle aeration (4 bubbles/sec) was maintained through a silicon rubber tube with a glass rod at the tip.

The hatching time (incubation duration) was determined when 50% of the eggs in each flask hatched. When the hatching finished, nauplii and eggs were collected with a 100-µm mesh and counted to determine the hatching rate.

Hatching rates were analyzed by two-way ANOVA. Differences were considered signifi-

cant at a 0.05 probability level by Scheffe's test after normality and homogeneity of the data were checked in the Minitab Statistical Package.

Results

Salinity and temperature and their interaction had significant influences on the hatching rate, but salinity appeared to have a more profound affect than temperature (Table 1). Eggs incubated at 40 and 35 ppt had higher ($p<0.05$) mean hatching rates (87.33-82.28%) than those incubated at 30 ppt (49.47%). In contrast to salinity, eggs incubated at the two higher temperatures had lower mean hatching rates than those incubated at the lowest temperature.

The eggs incubated at 24°C and 40 ppt had the best hatching rate ($p<0.05$). The lowest hatching rates were obtained at 30 ppt and 28°C or 32°C ($p<0.05$). Following hatching, larval activity was poor at 30 ppt regardless of the temperature.

Results indicate that successful egg incubation of *P. semisulcatus* can take place over a wide range of salinity and temperature, but the best salinity and temperature combination is 24°C and 40 ppt.

Discussion

The present study shows that salinity had a greater influence than temperature on the hatching rate of *P. semisulcatus*. The best hatching success was obtained at the highest salinity (40 ppt). Eggs incubated at 30 ppt had the lowest hatching rate. These findings agree with those of Nisa and Ahmed (2000) who studied the effects of six different salinities (20-45 ppt) on the eggs of four penaeid shrimps, *F. merguensis*, *F. penicillatus*, *M. affinis* and *P. stylifera*, in laboratory conditions. They stated that the best hatching success was at 35 ppt for all the species, hatching success decreased with the decrease in salinity, and no hatching occurred at 20-25 ppt for *P. stylifera*. Zacharia and Kakati (in press)

Table 1. Hatching rate (means \pm s.d., n= 3) of *Penaeus semisulcatus* eggs incubated in different salinity and temperature regimes.

Temperature (°C)	Salinity (ppt)	Hatching rate (%)	Hatching time (h)	Larval activity
24	30	84.00 \pm 7.11 ^a	17.5	Poor
	35	82.00 \pm 4.89 ^a	17.5	Good
	40	91.33 \pm 1.68 ^a	17.5	Good
28	30	18.67 \pm 9.29 ^{bc}	14.5	Poor
	35	81.00 \pm 5.68 ^a	14.5	Good
	40	84.17 \pm 9.05 ^a	14.5	Good
32	30	45.75 \pm 7.20 ^b	11.5	Poor
	35	83.83 \pm 7.14 ^a	11.5	Good
	40	86.50 \pm 0.98 ^a	11.5	Good

Means with different superscripts are significantly different ($p<0.05$).

also obtained the highest hatching rate at 33°C and 35 ppt, followed by 29°C and 35 ppt, in *F. merguiensis*. In the study by Preston (1985), both low (20 ppt) and high (40 ppt) salinities had a negative impact on hatching in *P. plebejus*, *M. macleayi* and *M. bennettiae*. On the other hand, Browdy and Samocha (1985) and Aktas and Kumlu (1999) reported that *P. semisulcatus* eggs incubated at 40 ppt had good hatching success, as demonstrated in the present study. Development of *F. indicus* and *P. semisulcatus* eggs was retarded at 20-25 ppt and eggs swelled to the point of bursting at 10-15 ppt (Tseng and Cheng, 1981, cited in Primavera, 1985). In addition, these researchers pointed out that *F. indicus* eggs had a significantly higher hatching rate and shorter incubation period at 33 ppt than at 22 ppt or 42 ppt. Our results and those reported in the literature indicate that the optimal egg incubation salinity for *P. semisulcatus* is about 40 ppt. Kumlu et al. (1999b and 2000) also reported this level of salinity as optimal for larviculture of this species.

Spawning and larval stages of most penaeid shrimp species occur in full strength sea water (>35 ppt) and at stable water temperatures. Hence, poorer hatching rates are normally expected in salinities lower than oceanic salinities. In the present study, 30 ppt salinity at all temperatures resulted in lower hatching rates and poorer larval activity after hatching. The best hatching success was achieved at the higher salinities of 35 and 40 ppt, regardless of the temperature. Similarly, among various temperature and salinity combinations, the highest mean hatching rate for *P. monodon* was obtained at 33 ppt and 23-33°C, whereas 23 and 28 ppt at any temperature produced weak larvae (Reyes 1981, cited in Primavera, 1985).

Incubation time was mainly affected by temperature in the present study, as suggested for *P. plebejus*, *Metapenaeus macleayi* and *M. bennettiae* by Preston (1985). Hatching rate peaked between 19 and 24°C for *P. plebejus* and between 24 and 29°C for *M. macleayi*. The optimal temperature for hatching *M. bennettiae* eggs was significantly affected by the conditions that existed at spawning (Lester and Pante,

1992). Yet, despite being spawned at 28°C, the hatching success of *P. semisulcatus* eggs was similar at all temperatures in the current study. Regardless of the salinity levels tested in our study, hatching occurred in 17.5 h at 24°C, while this duration was lower at 28°C and lower still at 32°C. Similar findings were reported for *P. monodon*, i.e., increasing the temperature from 23 to 33°C significantly decreased the incubation period (Primavera, 1985).

The results of the present study demonstrate that eggs of *P. semisulcatus* obtained from eastern Mediterranean broodstock should be incubated at 35-40 ppt and 24-32°C.

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